

Isolation Of Escherichia Coli And Klebsiella From Patient With Urinary Tract Infection

عزل بكتريا القولون المعوية والكلبيسيلا من المرضى المصابين بالتهاب المجاري البولية

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الخلاصة:

الخلفية: مقاومة بكتريا القولون المعوية والكلبيسيلا للمضادات الحيوية تسبب تعقيداً في معالجة التهاب المجاري البولية.
الهدف: ان الهدف من هذه الدراسة هو تقييم فعالية المضاد الحيوي ضد البكتريا المسببة لالتهاب المجاري (بكتريا القولون المعوية) البولية في محافظة النجف الاشرف.
طرق العمل: تم جمع ٨١ عينة ادرار من مرضى مصابين بالتهاب المجاري البولية وتم التحري عن بكتريا اشريشيا القولون والكلبيسيلا تم فحص حساسية العزلات للمضادات الحيوية.
النتائج: وجد أعلى معدل للمقاومة (١٠٠٪) للامبيسلين وأموكسيسيلين، بينما تكون المقاومة معتدلة للسيفالوتين.
الاستنتاج: وجد ان بكتريا القولون وبكتريا الكلبيسيلا متعددة المقاومة للمضاد الحيوي التي تم اختبارها في هذه الدراسة.

Abstract

Background. Resistance to antibiotics arising in *Escherichia coli* and *Klebsiella* species isolates may complicate therapeutic management of urinary tract infection(UTI) by this organism.

Aim. The aim of this study was to assess antibiotic activity against UTI isolates of *E.coli* in Al- Najaf governorate.

Methodology. A total of 81 mid –stream urine samples were collected from patient suspected of UTI and screened for the occurrence of *E.coli*.

Result. Susceptibility of the isolates to antibiotics was test by standard methods. Highest rate of resistance (100%) was found to ampicillin and amoxicillin, while moderately resistant to cephalothin .

Conclusion. The present study concluded that *E.coli* resistance to multiple antibiotic were recognized.

Keywords: Urinary tract infection, *Escherichia coli* , *Klebsiella*, Resistant to antibiotic.

INTRODUCTION

A UTI is defined as colonization of a pathogen occurring anywhere along the urinary tract: kidney, ureter, bladder, and urethra. Traditionally, UTIs have been classified by the site of infection (ie, pyelonephritis [kidney], cystitis [bladder], urethra [urethritis]) and by severity (ie, complicated versus uncomplicated). A complicated UTI describes infections in urinary tracts with structural or functional abnormalities or the presence of foreign objects, such as an indwelling urethral catheter. This model does not necessarily reflect clinical management, however. In children, a simpler and more practical approach is to categorize UTIs as a first infection versus recurrent infection. Recurrent infections can be further subdivided into unresolved bacteriuria, bacterial persistence, and reinfection.

The recurrence of a UTI may be caused by several reasons. Unresolved bacteriuria is most commonly caused by inadequate antimicrobial therapy. Subtherapeutic levels of the antimicrobial agents may be a result of noncompliance, malabsorption, suboptimal drug metabolism, and resistant uropathogens unresponsive to attempted therapy (1). In these cases, infection typically resolves after altering the therapy according to antimicrobial sensitivities determined by a proper urine culture. Bacterial persistence and reinfection occur after sterilization of the urine has been documented. In the case of bacterial persistence, the nidus of infection in the urinary tract is not eradicated. Characteristically, the same pathogen is documented on urine cultures during subsequent episodes of UTI despite negative cultures after treatment.

The uropathogen frequently resides in a location that is shielded from antimicrobial therapy. These protected sites are often anatomic abnormalities, including infected urinary calculi (2), necrotic papillus or foreign objects, such as an indwelling ureteral stent (3,4) or urethral catheters (5), which once infected may not be sterilized. Identification of the anatomic abnormality is essential because surgical intervention (extirpation) may be necessary to eradicate the source of infection.

E. coli is the most frequent documented uropathogen. Among neonates, UTI secondary to group B streptococci is more common than in older populations (6). In immune compromised children and children with indwelling catheters, *Candida* may be isolated from the urine. Nosocomial infections are typically more difficult to treat and are caused by various organisms, including *E. coli*, *Candida*, *Enterococcus*, *Enterobacter*, and *Pseudomonas* (7).

Bacterial clonal studies strongly support entry into the urinary tract by the fecal-perineal-urethral route with subsequent retrograde ascent into the bladder. Because of differences in anatomy, girls are at a higher risk of UTI than boys beyond the first year of life. In girls, the moist periurethral and vaginal area promotes the growth of uropathogens. The shorter urethral length increases the chance for ascending infection into the urinary tract. Once the uropathogen reaches the bladder, it may ascend to the ureters and then to the kidneys by some as-yet undefined mechanism. Additional pathways of infection include nosocomial infection through instrumentation, hematogenous seeding in the setting of systemic infection or a compromised immune system, and direct extension caused by the presence of fistulae from the bowel or vagina (8).

The urinary tract (ie, kidney, ureter, bladder, and urethra) is a closed, normally sterile space lined with mucosa composed of epithelium known as transitional cells. The main defense mechanism against UTI is constant antegrade flow of urine from the kidneys to the bladder with intermittent complete emptying of the bladder via the urethra. This washout effect of the urinary flow usually clears the urinary tract of pathogens. The urine itself also has specific antimicrobial characteristics, including low urine pH, polymorphonuclear cells, and Tamm-Horsfall glycoprotein, which inhibits bacterial adherence to the bladder mucosal wall (9).

UTI occurs when the introduction of pathogens into this space is associated with adherence to the mucosa of the urinary tract. If uropathogens are cleared inadequately by the washout effect of voiding, then microbial colonization potentially develops. Colonization may be followed by microbial multiplication and an associated inflammatory response. Bacteria that cause UTI in otherwise healthy hosts often exhibit distinctive properties known as virulence factors to overcome the normal defenses of the urinary system (10, 11). In serotypes of *E. coli* frequently isolated in UTI, bacterial adherence to the uroepithelium is enhanced by adhesins, often fimbriae (pili), which bind to specific receptors of the uroepithelium (12,13).

The interaction of fimbriae with the mucosal receptor triggers internalization of the bacterium into the epithelial cell, which leads to apoptosis, hyperinfection, and invasion into surrounding epithelial cells or establishment of a bacterial focus for recurrent UTI (14). Uropathogenic strains of *E. coli* have been recognized to release toxins, including cytotoxic necrotizing factor-1, secreted autotransporter toxin that causes cellular lysis, cause cell cycle arrest, and promote changes in cellular morphology and function (15,16). To promote survival, various uropathogens possess siderophore systems capable of acquiring iron, an essential bacterial micronutrient, from heme (17). Uropathogenic strains of *E. coli*

have a defensive mechanism that consists of a glycosylated polysaccharide capsule that interferes with phagocytosis and complement-mediated destruction.

The most common form of resistance to β -lactam agents is caused by enzymes that render molecules inactive by opening the β -lactam ring. In Gram-positive, β -lactamase is excreted into the medium and therefore destroys the antibiotics extracellularly (18). In contrast, the β -lactamase of Gram-negative bacteria is located in the periplasmic space where they attack the antibiotic before it can reach its receptor site.

METHODOLOGY:

Present study was carried out in two hospitals in Najaf (Al-Sadder Teaching and Al-Zahra Maternity and children). During the period from July to October 2012, a total of 100 urine samples collected (by mid – stream urine) from patients suspected to have UTI were culture on blood agar (Hi-media Laboratories, India) and CHROM agar (Oxoid, France) using standard method. Gram negative rod isolated in significant counts ($>10^5$ cfu/ml) in pure culture were included in the study. Identification of isolates up to the species level was done according to conventional scheme of MacFaddin (19) and was confirmed by an additional biochemical test with API20E miniaturized diagnostic test (Bio Merieux, France).

RESULTS

The present study included a collection of 100 urine samples from two hospitals in Najaf during the period from July to October, 2012. Out of the 100 samples processed; 81 (81%) showed significant bacteriuria. A total of 55 (67.9%) females and 26 (32%) males had positive urine culture (significant bacteriuria). The bacterial isolates obtained as a pure and predominant growth from urine samples were only considered for the present study.

All bacteria isolated in this study were identified based on colonial morphology, and biochemical reactions according to MacFaddin (2000). The organisms grown on the culture of all the 81 urine samples with significant bacteriuria were as follows: 43 (53%) isolates of *E. coli*, 26 (32%) isolates of *Klebsiella* spp., 8 (9.8%) isolates of *Proteus* spp., and 4 (4.9%) isolates of *Pseudomonas* spp. The frequency of antibiotic resistance of the 43 *E. coli* isolates. *E. coli* isolates (100%) were found to be resistant to ampicillin and amoxicillin. (94.7%) of *E. coli* isolates were highly resistant to oxacillin, and moderately resistant to cephalothin (22.5%), ciprofloxacin, gentamycin, trimethoprim (46.5%), nitrofurantoin (44.1%), nalidixic acid (37.2%), cefotaxime (39.5%), ceftazidime (34.8%), and chloramphenicol (27.9%). On the other hand, all *E. coli* isolates were susceptible to imipenem.

DISCUSSION

Urinary tract infections are the most common type of the clinical disease produced by the *E. coli*. The results showed a high incidence of UTI in females than males, which might be due to variety of factors, such as the close proximity of the female urethral meatus to the anus (20), and alternations in vaginal microflora that play a critical role in encouraging vaginal colonization with coliforms which may lead to UTI (21, 22). (23) Reported that the bladder infections are 14-times more common in females than males by virtue of the shortened urethra. Moreover, (24) showed that UTI are more common in females, 40% of women have an episode in their lifetime

when they are sexually active.

Numerous in vitro studies have shown the *E. coli* and *Klebsiella* spp. as the most common causes of UTI (25). All *Klebsiella* isolates were identified according to, The development of antibiotics resistance in these isolates is often related to the overuse and misuse of the antibiotics prescribed. Iraq is one of the developing countries where antibiotics sold over the counter, an attitude that encourages self-medication.

In this investigation, the reason of β -lactam resistance of *E. coli* isolates is probably due to the production of TEM β -lactamases, which may be genetically localized on the chromosome or on a plasmid. The TEM-1 is the most commonly encountered β -lactamase in Gram-negative bacteria; up to 90% of ampicillin resistance in *E. coli* is due to the production of TEM-1 (26). Other studies from many areas reported an increasing resistance of *E. coli* to ampicillin.

In this investigation, although the β -lactamases undoubtedly play a major role in the resistance to β -lactam antibiotics, the high ratio of resistance to ampicillin and amoxicillin was not only attributable to the production of β -lactamase enzymes. The other mechanisms conferring resistance to these compounds is caused by reducing of the activity of β -lactam antibiotics in a resistant cell due to many factors such as; the sensitivity of the antibiotic to β -lactamases, the penetration through the outer membrane, the affinity for the target (PBPs), the amount of β -lactamase, and the affinity of the antibiotic for the β -lactamase . A range of antibiotics have been used for the treatment of UTI caused by *E. coli* and *K.pneumoniae* in Iraq and other countries. However, the widely spread use of this approach has criticized on the ground of drug toxicity and the risk of an increase spread of antibiotic resistance (27).

Bacterial resistance to antibiotics is now widespread and possesses serious clinical threats. The organisms develop resistance to antibiotics by any of the following mechanisms: selection, mutation, phage transduction, and transference. Microbial resistance can be either hereditary in the organism or acquired through the environment. The high resistance in the present study may be due to antibiotic abuse which leads to development of resistant isolates in Iraq. In agreement with the present study, (28) found that 56.8% of clinical *E. coli* isolates in Najaf were resistant to more than five antimicrobial agents.

CONCLUSION:

The study concluded *Escherichia coli* and *K. pneumoniae* were the predominant species recovered in patients with significant bacteriuria. Most of the test isolates were resistant to antibiotics, for that reason such organism pose a serious therapeutic problem in Najaf hospitals.

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